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**First records of *Hyalomma rufipes* and *Ixodes neitzi* (Acari: Ixodidae) found on large
carnivores in South Africa**

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Abstract

Ixodid ticks (Acari: Ixodidae) are important disease vectors for large carnivores, but the composition of the tick communities that parasitize carnivores is poorly understood. We collected ticks from leopards (*Panthera pardus*) and brown hyenas (*Hyaena brunnea*) in the Soutpansberg Mountains, South Africa, to determine which species feed on these carnivores. We identified a total of eight tick species belonging to six genera, and recorded *Ixodes neitzi* and *Hyalomma rufipes* on *P. pardus* for the first time.

Keywords: *Ixodes neitzi*, *Hyalomma rufipes*, leopard, brown hyena

1. Introduction

Ticks are among the most important vectors of pathogens of wildlife worldwide (Pfäffle et al., 2013; Jongejan and Uilenberg, 2004). The impact of tick-borne diseases is of increasing concern to large carnivores globally, as populations are often threatened or significantly declining (Ripple et al., 2014). Disease can be an important cause of carnivore mortality (Murray et al. 1999), leading to population declines (Trinkel and Angelici, 2016) and extirpation (Cleaveland et al., 2007), with associated cascading effects on communities (Hollings et al., 2014). Parasite infection can also have substantial sub-lethal effects on hosts, including reducing foraging efficiency and competitive ability (Barber et al. 2000), and lowering reproductive success (Simmons and Zuk, 1992).

To date, very few studies of ticks have been conducted on large predators in Africa, and only a handful of individual leopards (*Panthera pardus*) and brown hyenas (*Hyaena brunnea*) have been sampled for ticks (Table A.1, Theiler 1962 in Boomker et al. 1997, I.G. Horak unpublished data in Boomker et al. 1997, Horak et al. 2000, Horak et al. 2010, Walker et al. 2000), mostly due to the difficulty of sampling these wild animals. This study adds observations to the current knowledge of tick species hosted by wild carnivores by cataloguing the tick species present on *P. pardus* and *H. brunnea* in the Soutpansberg Mountains, South Africa.

2. Methods

The study focussed on the Luvhondo Private Nature Reserve and neighbouring properties,

which are located in the western Soutpansberg Mountain Range, Limpopo Province, South Africa (29°26'E, 23°01'S, altitude ca. 1,420 m), an area recognised as a biodiversity hotspot (Foord et al., 2002). The large carnivore guild resident in the area consists of *P. pardus* and *H. brunnea*. Although local population density estimates of *P. pardus* were previously high (Chase Grey et al., 2013), they appear to be declining (Williams et al., 2017), while those of *H. brunnea* are comparable to those recorded elsewhere (Williams, 2017).

Six *P. pardus* individuals and four *H. brunnea* individuals were live-trapped and immobilized as part of other studies on predator ecology (Williams, 2017; Williams et al., 2017) between June 2012 and October 2013 (Table A.2), using soft-hold foot traps (Frank et al., 2003).

Details of capture procedures are provided in Williams (2017) and Williams et al. (2017).

Ticks were opportunistically collected from animals by hand for up to 30 minutes, the maximum time possible before the animals started to wake up. All encountered life stages of ticks were collected (as reflected in Table 1). Ticks were preserved in vials containing 70% ethanol and stored at room temperature. Identification of the collected ticks was conducted by HH at the Onderstepoort Veterinary Institute, Pretoria, South Africa (references for identification: Apanaskevich and Horak, 2008; Apanaskevich et al., 2007; Clifford et al., 1977; Robinson, 1926; Theiler, 1961; Voltzit and Keirans, 2003; Walker and Olwage, 1987; Walker et al., 2000).

3. Results and discussion

Eight tick species belonging to six genera were identified from *P. pardus* (6 animals, 67 ticks)

and *H. brunnea* (4 animals, 113 ticks) in the western Soutpansberg Mountains, including the first recorded cases of *Hyalomma rufipes* and *Ixodes neitzi* occurring on *P. pardus* (adult ticks). *Amblyomma hebraeum*, *Haemaphysalis elliptica*, *Rhipicentor nuttalli* and *Rhipicephalus simus* were found on both carnivore species. *Rhipicephalus appendiculatus* and *Rhipicephalus zambeziensis* were collected from *H. brunnea* only (Table 1).

H. rufipes is a two-host tick, which spends its first two parasitic life stages, larva and nymph, on a single avian, leporid or rodent host and its adult stage on a large mammalian host (Dransfield and Brightfield, 2013). Since this tick species is not known to be associated with wild carnivores, and we collected only a single specimen from a single *P. pardus*, this may be an incidental finding. In contrast, *I. neitzi* was found on three out of six *P. pardus* individuals sampled. The species has been mentioned in scientific literature only a few times. The first account, and most other records (14 animals infested), were associated with mountain reedbuck (*Redunca fulvorufula*) (Horak et al., 2018, Clifford et al., 1977). Besides occasional collections from a klipspringer (*Oreotragus oreotragus*) (Rechav et al., 1978), a greater kudu (*Tragelaphus strepsiceros*), and an eland (*Tragelaphus oryx*) (Horak et al., 2018), our results represent the first account for large carnivores. Other than these incidental collections, however, host association and life cycle characteristics are unknown (Horak et al., 2018). While adults have been described as most active from late spring to summer (Clifford et al., 1977), it is interesting to note that two of our three samples were collected in winter (June and July) with only one from summer (December).

We notice a much larger number of tick species on *P. pardus* (52, this study included) compared to *H. brunnea* (9) (Table A1). This may in part reflect sampling efforts on the two

species. However, there is no real evidence of more intense screening for ticks on *P. pardus* compared to *H. brunnea* in the literature, and, to our knowledge, there are no published reports on the number of *P. pardus* compared to the number of *H. brunnea* being caught. Compared to *H. brunnea*, *P. pardus* inhabits a wider variety of habitat, and is, accordingly, exposed to a wider diversity of tick species (Horak et al., 2018; Skinner and Smithers, 1990). *H. brunnea* is associated with the dryer parts of the southern savanna (Skinner and Smithers, 1990), while many of the tick species collected from leopards (Table A1), especially the *Ixodes* species, occur in more humid habitats. Whether habitat use differences or host- and/or tick-specific characteristics are at the basis of this contrast remains to be studied.

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Table 1 Overview of collected ticks from *P. pardus* and *H. brunnea* in this study.

Tick Species	Host species	
	<i>P. pardus</i>	<i>H. brunnea</i>
<i>Amblyomma hebraeum</i>	1L 1♂ (2)	1L 8N 1♂ (3)
<i>Haemaphysalis elliptica</i>	1♀ (1)	3♂ 5♀ (3)
<i>Hyalomma rufipes</i>	1♂ (1)	
<i>Ixodes neitzi</i>	4♀ (3)	
<i>Rhipicentor nuttalli</i>	32♂ 9♀ (6)	14♀ (2)
<i>Rhipicephalus appendiculatus</i>		26♂ 35♀ (2)
<i>Rhipicephalus simus</i>	10♂ 8♀ (1)	5♂ 14♀ (2)
<i>Rhipicephalus zambeziensis</i>		1♀ (1)
No. of tick species	6 (total number of ticks: 67)	6 (total number of ticks: 113)

Collected between June 2012 and October 2013 in the Soutpansberg Mountains, South Africa. Types of ticks found are indicated by L (larvae), N (nymphs), ♂ (adult males), and ♀ (adult females). Numbers in brackets refer to the number of individual animals from which those ticks were collected. Tick species marked in bold indicate tick species newly found on the respective host species.

Table A.1 List of tick species recorded for leopards (*Panthera pardus*) and brown hyaena (*Hyaena brunnea*) in Africa.

Tick species	<i>P. pardus</i>	<i>H. brunnea</i>
<i>Amblyomma hebraeum</i>	A, C, D, E	E
<i>Amblyomma marmoreum</i>	E	
<i>Amblyomma nuttalli</i>	A	
<i>Amblyomma tholloni</i>	A	
<i>Amblyomma variegatum</i>	A	
<i>Haemaphysalis aciculifer</i>	A	
<i>Haemaphysalis elliptica</i>	A*, C*, D, E	C, E
<i>Haemaphysalis leachi/zumpti</i>	C, E	
<i>Haemaphysalis parvula</i>	A	
<i>Hyalomma truncatum</i>	A	
<i>Ixodes cavipalpus</i>	A	
<i>Ixodes cumulatimpunctatus</i>	A	
<i>Ixodes moreli</i>	A	
<i>Ixodes muniensis</i>	A	
<i>Ixodes oldi</i>	A	
<i>Ixodes pilosus</i>	A	
<i>Ixodes rasilus</i>	A	
<i>Ixodes vanidicus</i>	A	
<i>Rhipicentor bicornis</i>	A, D	
<i>Rhipicentor nuttalli</i>	C, D, E	C, E
<i>Rhipicentor sp.</i>	A	
<i>Rhipicephalus appendiculatus</i>	A, C, E, F	F
<i>Rhipicephalus aquatilis</i>	F	
<i>Rhipicephalus armatus</i>	A, F	

<i>Rhipicephalus capensis</i>	A, D, E	
<i>Rhipicephalus compositus</i>	A	
<i>Rhipicephalus cuspidatus</i>	F	
<i>Rhipicephalus decoloratus</i>	E	
<i>Rhipicephalus evertsi evertsi</i>	B, D, F**	
<i>Rhipicephalus evertsi</i> <i>mimeticus</i>	F	
<i>Rhipicephalus gertrudae</i>	F	
<i>Rhipicephalus hurti</i>	F	
<i>Rhipicephalus kochi</i>	F	
<i>Rhipicephalus longus</i>	F	
<i>Rhipicephalus lunulatus</i>	F	
<i>Rhipicephalus masseyi</i>	F	
<i>Rhipicephalus muhsamae</i>	F	
<i>Rhipicephalus praetextatus</i>	F	
<i>Rhipicephalus pravus</i>	A	
<i>Rhipicephalus pulchellus</i>	A, F	
<i>Rhipicephalus</i> (nr.) <i>punctatus</i> ***	F	
<i>Rhipicephalus sanguineus</i> <i>sensu lato</i>	A****	F
<i>Rhipicephalus senegalensis</i>	A	
<i>Rhipicephalus serranoi</i>	F	
<i>Rhipicephalus simus</i>	A, C, E, F	C, E, F
<i>Rhipicephalus sulcatus</i>	A	E
<i>Rhipicephalus tricuspis</i>	A, F	
<i>Rhipicephalus turanicus</i>	B, C, E, F	F
<i>Rhipicephalus zambeziensis</i>	B, C, D, E, F	F
<i>Rhipicephalus ziemanni</i>	A, F	
No. of tick species	50	9

By (A) Theiler 1962 in Boomker et al. 1997 (Africa), (B) I.G. Horak unpublished data in Boomker et al. 1997 (South Africa), (C) Horak et al. 2000 (South Africa), (D) Horak et al. 2010 (Botswana, South Africa, Namibia), (E) Horak et al. 2018 (Southern Africa) and (F) Walker et al. 2000 (only tick species occurring in the Afrotropical region are included). Data in (F) is a combination of earlier published studies, and (at that time) unpublished data, and may thus in part be the same observations as the observations from A-D. Many observations in (E) are occasional ones. * Historically *Ha. colesbergensis* and *Ha. elliptica* specimens in South Africa were misidentified as *Haemaphysalis leachi*. The historical *Ha. elliptica* is now regarded as three different species (*Ha. elliptica* and *Ha. colesbergensis* in South Africa and *Ha. leachi* not occurring in South Africa). The specimens from A and C were originally misidentified and reported as *Haemaphysalis laeichi* (Apanaskevich et al. 2007). ** Should be considered accidental (Walker et al. 2000 p.169). *** Ticks were identified as *Rhipicephalus* near *punctatus*. This species in South Africa (also reported as *Rhipicephalus punctatus* group, *Rhipicephalus* (nr.) *pravus*, *Rhipicephalus pravus* group, *Rhipicephalus* sp. near *pravus* or *Rhipicephalus* nr. *warburtoni*) is now, after careful studies of all stages of the tick species, considered to be *Rhipicephalus warburtoni sensu stricto* (I.G. Horak personal communication). **** Probably refers to *R. turanicus* (Walker et al. 2000 pp.387-388 & 458, Pegram et al. 1987).

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Table A.2 Capture dates and sex for the host species sampled.

Species	Date caught	Sex
Leopard (<i>Panthera pardus</i>)	June 2012	M
	June 2012	M
	July 2012	F
	February 2013	M
	June 2013	M*
	September 2013	M*
	September 2013	F
Brown hyaena (<i>Hyaena brunnea</i>)	February 2013	F
	March 2013	M

	September 2013	F
	October 2013	F

*The same individual was caught twice.

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